

We claim:.

1. A non-radioactive hybridization assay for the detection of a target nucleic acid sequence in a biological sample the improvement comprising the steps of:
 - a) hydrolyzing the sample with a base;
 - b) hybridizing a nucleic acid sequence in the treated sample to a complementary nucleic acid probe to form a double-stranded hybrid;
 - c) capturing the hybrid onto a solid phase to which an anti-hybrid antibody or anti-hybrid fragment has been immobilized; and,
 - d) eliminating any non-hybridized probe; and
 - e) detecting the bound hybrid.
2. The assay of claim 1 wherein the non-hybridized probe is eliminated by digestion with an enzyme.
3. The assay of claim 1 wherein the target nucleic acid is DNA selected from the group consisting of, human papillomavirus DNA, hepatitis B DNA, and *Chlamydia* DNA.
4. The assay of claim 1 wherein the probe is an RNA sequence complementary to target DNA.
5. The assay of claim 1 wherein the double-stranded hybrid is an RNA/DNA hybrid.
6. The assay of claim 1 wherein the digesting enzyme is RNAase.
7. The assay of claim 1 wherein the concentration of probe is between 1 and 500 ng/ml.
8. The assay of claim 1 wherein the concentration of probe is between 20 and 200 ng/ml.
9. The assay of claim 1 wherein the concentration of probe is approximately 75 ng/ml.
10. The assay of claim 1 wherein the base is sodium hydroxide in a concentration of between 0.1 and 2.0 M, incubated with the sample at a temperature between 20 and 100°C for a period of between 5 and 120 minutes.

11. The assay of claim 1 wherein the base is sodium hydroxide in a concentration of between 0.2 and 0.8 M, incubated with the sample at a temperature between 60 and 70°C for a period of between 30 and 60 minutes.

12. The assay of claim 1 wherein the base is sodium hydroxide in a concentration of approximately 0.415 M, incubated with the sample at a temperature of approximately 65°C for a period of approximately 45 minutes.

13. The assay of claim 6 wherein the RNAase is added to the sample in a concentration between 0.01 and 1 mg/ml and incubated with the sample at a temperature between 4 and 45°C for a period of between 5 minutes and 24 hours.

14. The assay of claim 6 wherein the RNAase is added to the sample in a concentration between 0.05 and 0.5 mg/ml and incubated with the sample at a temperature between 20 and 30°C for a period of between 10 and 60 minutes.

15. The assay of claim 6 wherein the RNAase is added to the sample in a concentration of approximately 0.2 mg/ml and incubated with the sample at room temperature for a period of approximately 30 minutes.

16. The assay of claim 1 further comprising diluting the probe in a buffer that restores the sample to a neutral pH.

17. The assay of claim 16 wherein the buffer comprises 2-[bis(2-Hydroxyethyl) amino] ethane sulfonic acid and sodium acetate.

18. A kit for the detection of a target nucleic acid sequence for diagnosing genetic defects, microbial or viral infections in a biological sample comprising:

- a) a sample transport medium for stabilization of the biological sample;
- b) a base for treating the sample by nicking and degrading the target nucleic acid sequence therein;
- c) a probe complementary to the treated target nucleic acid sequence for formation of a double-stranded nucleic acid hybrid;

- d) a neutralizing probe diluent for diluting the probe and neutralizing the treated target nucleic acid sequence;
- e) a solid phase to which an anti-hybrid antibody or an anti-hybrid antibody fragment has been immobilized, wherein the antibody is specific for a hybrid formed by hybridization of the probe and the target nucleic acid sequence;
- f) means for eliminating any non-hybridized probe; and,
- g) means for detecting the hybrid formed by hybridization of the probe and the target nucleic acid sequence.

19. The kit of claim 18 wherein the means for eliminating non-hybridized probe is an enzyme that digests non-hybridized probe.

20. The kit of claim 18 wherein the target nucleic acid is DNA selected from the group consisting of human papillomavirus DNA, hepatitis B virus DNA and *Chlamydia* DNA.

21. The kit of claim 18 wherein the probe is an RNA sequence complementary to target DNA.

22. The kit of claim 18 wherein the digesting enzyme is RNAase.

23. The kit of claim 18 wherein the base is sodium hydroxide in a concentration of between 0.1 and 2 M.

24. The kit of claim 18 wherein the digesting enzyme and detecting means are combined in a single reagent.

25. A buffer for use in a hybridization assay comprising 2-[bis(2-Hydroxyethyl) amino] ethane sulfonic acid and sodium acetate wherein the pH of the buffer is between approximately 5 and 5.5.

26. The buffer of claim 25 further comprising an antimicrobial agent, a metal chelating agent, a detergent, and a hybridization accelerator.

27. The buffer of claim 25 further comprising a probe in a concentration between 1 and 500 ng/ml for hybridization to a target nucleic acid.

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